REMARKS

Claims 124, 137, 139, and 150 have been amended. New claims 151-168 have been added. Claims 128 and 143 have been canceled. Claims 124-127 and 128-168 are pending in the present application.

Claims 124-150 are pending in the present application. It is respectfully submitted that the present amendment presents no new issues or new matter and places this case in condition for allowance. Reconsideration of the application in view of the above amendments and the following remarks is requested.

I. Objections to Claims 143 and 150

Claims 143 and 150 are objected to due to the recitation of "a third nucleic acid". The Office Action suggested that the term be replaced with "the third nucleic acid" since the nucleic acid has been already defined in the claims from which claims 143 and 150 depend. Applicant respectfully points out that the term "a third nucleic acid" appears in claims 137 and 150, not claim 143.

Claims 137 and 150 were amended to recite "the third nucleic acid",

Applicant submits that the objections have been overcome and respectfully request withdrawal of the objections.

II. Rejection of Claims 124-127, 131-142, and 146-150 under 35 U.S.C. § 112, First Paragraph

Claims 124-127, 131-142, and 146-150 remain rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the written description requirement for reasons of record. The Office Action states:

> The claimed invention requires the disruption or deletion of a genus of Fusarium venenatum cyclohexadepsipeptide synthetase genes. As such, one of skill in the art would require knowledge or guidance as to which is the structure of the gene to be disrupted or which elements of said genes are to be deleted to eliminate function. Furthermore, while the structure of the polynucleotide of SEQ ID NO: 1 is known, there is no teaching in the specification which indicates how the structure of the polynucleotide of SEQ ID NO: 1 correlates with the structure of any *F. venenatum* cyclohexadepsipeptide synthetase gene, i.e., degree of structural similarity among all *F. venenatum* cyclohexadepsipeptide synthetase genes. No disclosure of the critical structural elements in the polynucleotide of SEQ ID NO: 1 (or the polynucleotide used by Herrmann *et al.*) that correlate with

> cyclohexadepsipeptide synthetase function which are common to all *F. venenatum* cyclohexadepsipeptide synthetase genes has been presented, such that, for example, one of skill in the art would know which fragments of the polynucleotide of SEQ ID NO: 1 (or the polynucleotide used by Herrmann *et al.*) can be used in homologous recombination to inactivate all *F. venenatum* cyclohexadepsipeptide synthetase genes.

The rejections are respectfully traversed.

The present invention is directed to methods for producing a secreted heterologous polypeptide, comprising: (a) cultivating a mutant cell of a parent *Fusarium venenatum* cell under conditions conducive for the production of the secreted heterologous polypeptide, wherein (i) the mutant cell comprises a first nucleic acid encoding the secreted heterologous polypeptide, and (ii) the mutant cell comprises a second nucleic acid which comprises a disruption or a deletion in a cyclohexadepsipeptide synthetase gene, wherein the mutant cell produces less cyclohexadepsipeptide than the parent *Fusarium venenatum* cell when cultured under the same conditions as a result of the disruption or the deletion in the cyclohexadepsipeptide synthetase gene, wherein the cyclohexadepsipeptide synthetase gene encodes a cyclohexadepsipeptide synthetase having an amino acid sequence which has at least 70% identity with SEQ ID NO: 2; or a cyclohexadepsipeptide synthetase which is encoded by a nucleic acid which hybridizes under at least medium stringency conditions with (i) the nucleic acid of SEQ ID NO: 1, (ii) the cDNA of SEQ ID NO: 1, or (iii) a complete complementary strand of (i) or (ii), wherein medium stringency conditions are defined as prehybridization and hybridization at 45°C in 5X SSPE, 0.3% SDS, 200 μg/ml sheared and denatured salmon sperm DNA, and 35% formamide and washing three times each for 15 minutes using 2X SSC, 0.2% SDS at 55°C; and (b) isolating the secreted heterologous polypeptide from the cultivation medium.

Applicant submits that the specification complies with the written description requirement.

It is well settled that "[t]he test for determining compliance with the written description requirement is whether the disclosure of the application as originally filed reasonably conveys to the artisan that the inventor had possession at that time of the later claimed subject matter ..." *In re Kaslow*, 217 USPQ 1089, 1096 (Fed. Cir. 1983).

As set forth in Federal Circuit decisions, a specification complies with the written description requirement if it provides "a precise definition, such as by structure, formula, chemical name, or physical properties of the claimed subject matter sufficient to distinguish it from other materials." See, e.g., *University of California v. Eli Lilly and Co.*, 43 U.S.P.Q.2d 1398, 1404 (Fed. Cir. 1997); *Enzo Biochem v. Gen-Probe Inc.*, 63 U.S.P.Q.2d 1609, 1613 (Fed. Cir. 2002). In fact, "[a]

description of a genus of cDNAs may be achieved by means of a recitation of a representative number of cDNAs, defined by nucleotide sequence, falling within the scope of the genus or of a recitation of structural features common to the members of the genus, which features constitute a substantial portion of the genus." *Eli Lilly and Co.*, 43 U.S.P.Q.2d at 1569.

The Federal Circuit provides that the written description requirement for a genus of DNAs is met by a recitation of a representative number of DNAs, defined by nucleotide sequence, falling within the scope of the genus or by a recitation of structural features common to the members of the genus.

It is well established in the art that the definition of a genus of genes encoding polypeptides having an enzyme activity of interest is accomplished by using structural features that show the relatedness of the genes and their encoded products. For decades the scientific community has employed three structural features to define the relatedness of genes and their products. The three structural features are (1) percent identity of the amino acid sequences encoded by the genes, (2) percent homology of the nucleic acid sequences of the genes, and (3) nucleic acid hybridizations under defined stringent conditions to identify complementary strands of genes encoding the same or similar enzyme or protein function. These structural features have been used to predict the function of polypeptides encoded by novel genes, and to place them in an existing genus.

A search of the protein databases failed to reveal any prior art cyclohexadepsipeptide synthetase that shares at least 70% identity with the cyclohexadepsipeptide synthetase of SEQ ID NO: 2 of the present invention. Thus, the cyclohexadepsipeptide synthetase of the instant invention (SEQ ID NO: 2) is a new genus. Limiting the literal scope of protection of such a new genus or family to the nucleic acid sequence of SEQ ID NO: 1 or the amino acid sequence of SEQ ID NO: 2 provides little incentive to an Applicant to seek patent protection because biological diversity dictates that there will be natural variation in the sequences of other homologous genes existing in nature that are structurally- and functionally-related. Biological diversity in a given gene sequence can easily be found. As genes that fulfill a similar function in different species have evolved from a common ancestor, natural variation in the nucleic acid sequence will rapidly evolve following this speciation. Sequence variation within a single species is also common.

In the claims at issue, Applicant provides a recitation of two structural features common to the claimed genus: (1) the cyclohexadepsipeptide synthetase gene encodes a cyclohexadepsipeptide synthetase having an amino acid sequence which has at least 70%

identity with SEQ ID NO: 2; and (2) a cyclohexadepsipeptide synthetase which is encoded by a nucleic acid which hybridizes under at least medium stringency conditions with (i) the nucleic acid of SEQ ID NO: 1, (ii) the cDNA of SEQ ID NO: 1, or (iii) a complete complementary strand of (i) or (ii), wherein medium stringency conditions are defined as prehybridization and hybridization at 45°C in 5X SSPE, 0.3% SDS, 200 μg/ml sheared and denatured salmon sperm DNA, and 35% formamide and washing three times each for 15 minutes using 2X SSC, 0.2% SDS at 55°C.

The Office states that while the structure of the polynucleotide of SEQ ID NO: 1 is known, there is no teaching in the specification which indicates how the structure of the polynucleotide of SEQ ID NO: 1 correlates with the structure of any *F. venenatum* cyclohexadepsipeptide synthetase gene. With due respect, this statement is without merit. The structural features are described on page 18, line 8, to page 22, line 27, of the specification. As mentioned above, the structural features of percent identity at the deduced amino acid sequence level and the ability of the claimed nucleic acid sequence to hybridize under specific stringency conditions have been used for decades by persons of ordinary skill in the art to determine the relatedness of proteins and their genes with respect to structure and function to ascertain whether they belong to the same genus or family. The scientific literature abounds with disclosures of these structural features to describe the relatedness of proteins and their genes as well as to distinguish a protein and its gene from other proteins and their genes. Moreover, annotated databases of families of structurally-related proteins with a specific biological activity have been constructed based on these structural features. For example, the CAZy database describes the families of structurally-related catalytic and carbohydrate-binding modules (or functional domains) of enzymes that degrade, modify, or create glycosidic bonds. See www.afmb.cnrs-mrs.fr/CAZY/.

The Office contends that the structural features of percent identity at the deduced amino acid sequence level and the ability of the nucleic acid sequence to hybridize under specific stringency conditions are arbitrary since neither the specification nor the prior art discloses any definitive relationship between protein function and % identity or homology at either the nucleotide or amino acid level. With due respect, this statement is without merit. It is well established in the art that there is a definitive relationship between protein function and % identity or homology at either the nucleotide or amino acid level. Percent identity is highly predictive of protein function and without this tool it would be impossible to make meaningful annotations of genomes in sequencing projects. Proteins that share 70% amino acid identity are

known to possess the same catalytic/biochemical function which has formed the basis for genome annotation and comparative genomics. In fact, 70% identity is an extremely conservative criterion for judging functional similarity. A long history of structure-function studies has demonstrated that single domain proteins that share substantial similarity (and >30% identity) over their entire length (>80 residues) without introduction of numerous gaps are almost certainly homologous (derive from a common evolutionary ancestor) and share the same three-dimensional structure (see Marti´-Renom MA, Stuart AC, Fiser A, Sanchez R, Melo F, Sali A. Comparative protein structure modeling of genes and genomes. *Annu. Rev. Biophys. Biomol. Struct.* 2000; 29:291–325). At the 70% level of amino acid identity, orthologous enzymes in related species are virtually guaranteed to share the same catalytic function and substrate specificity. A simple search of any public database using the criteria above for a reference protein of interest will prove that there is a definitive relationship between protein function and % identity or homology at either the nucleotide or amino acid level.

In fact, the PTO has determined that the written description requirement can be met by "show[ing] that an invention is complete by disclosure of sufficiently detailed, relevant identifying characteristics ... *i.e.,* complete or partial structure, other physical and/or chemical properties, *functional characteristics when coupled with a known or disclosed correlation between function and structure,* or some combination of such characteristics." Guidelines for Examination of Patent Applications Under the 35 U.S.C. 112, ¶ 1 "Written Description" Requirement, 66 *Fed.Reg.* 1099, 1106 (Jan. 5, 2001).

In the claims at issue, each of the claimed structural features (percent identity and hybridization) specifies a family of structurally- and functionally-related enzymes having cyclohexadepsipeptide synthetase activity. Since the claimed structural features provide a correlation between function and structure, the written description requirement is satisfied.

The Office Action also suggests that one of skill in the art would not know which fragments of the polynucleotide of SEQ ID NO: 1 (or the polynucleotide used by Herrmann *et al.*) can be used in homologous recombination to inactivate all *F. venenatum* cyclohexadepsipeptide synthetase genes. With due respect, this statement is without merit. Applicant has shown that the deduced amino acid sequence (SEQ ID NO: 2) of the cyclohexadepsipeptide synthetase gene of SEQ ID NO: 1 shares approximately 59% identity with the deduced amino acid sequence of the cyclohexadepsipeptide synthetase gene (*esyn1*) of *Fusarium scirpi* (Haese *et al.*, 1993, *Mol. Microbiol.* 7: 905-914; DNA sequence listed in EMBL database under accession number Z18755). This sequence comparison indicated there are

regions of conserved homology between the sequences at the DNA level, which can be used to construct a disruption or deletion vector for use in another *Fusarium venenatum* cell without any knowledge of the DNA sequence in that cell. Applicant has provided evidence of this state of art by reference to Herrmann *et al.* (*Molecular Plant-Microbe Interactions* 9: 226-232, 1996) who showed that an internal fragment of the *Fusarium scirpi* cyclohexadepsipeptide synthetase gene was useful in disrupting the *Fusarium avenaceum* cyclohexadepsipeptide synthetase gene without any knowledge of the full nucleic acid sequence of the *Fusarium avenaceum* gene. Consequently, Applicant's disclosure combined with the knowledge in the prior art can be used to construct a mutant cell of a *Fusarium venenatum* parent strain, wherein the mutant comprises a disruption or a deletion in a cyclohexadepsipeptide synthetase gene, wherein the mutant cell produces less cyclohexadepsipeptide than the parent *Fusarium venenatum* cell when cultured under the same conditions as a result of the disruption or the deletion in the cyclohexadepsipeptide synthetase gene.

For the foregoing reasons, Applicant submits that the new claims overcome the rejections under 35 U.S.C. § 112, first paragraph. Applicant respectfully requests reconsideration and withdrawal of the rejection.

III. Rejection of Claims 124-128, 131-143, and 146-150 under 35 U.S.C. § 112, First Paragraph

Claims 124-128, 131-143, and 146-150 remain rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement for reasons of record The Office Action states:

> It is reiterated herein that one of skill in the art would require, at the very least. some knowledge or guidance as to how the structure of the polynucleotide of SEQ ID NO: 1 correlates with the structure of all *F. venenatum* cyclohexadepsipeptide synthetase genes in order to know which polynucleotide has to be inserted in a disruption or deletion vectors such that upon transformation, disruption or deletion of any *F. venenatum* cyclohexadepsipeptide synthetase gene would occur. As indicated above, neither the specification nor the art teaches which are the structural elements associated with cyclohexadepsipeptide synthetase function in the polynucleotide of SEQ ID NO: 1 (or the polynucleotide used by Herrmann *et al.*) that are common in *F. venenatum* cyclohexadepsipeptide synthetase genes. In addition, there is no disclosure of the degree of structural similarity shared among all the *F. venenatum* cyclohexadepsipeptide synthetase genes and the only structure disclosed, i.e. SEQ ID NO: 1. No disclosure has been provided as to the structural elements in the polynucleotide of SEQ ID NO: 1 which can be deleted, substituted or inserted to create a structural homolog encoding a polypeptide having 70%

sequence identity to the polypeptide of SEQ ID NO: 2 with the same function as that of the polypeptide of SEQ ID NO: 2. Similarly, no disclosure has been provided of the structural elements in the polynucleotide of SEQ ID NO: 1 which must be present in a polynucleotide hybridizing under medium stringency conditions as recited such that it encodes a polypeptide having the same function as that of the polypeptide of SEQ ID NO: 2. Therefore, due to the lack of relevant examples, the amount of information provided, the lack of knowledge as to the structmal elements required in all *F. venenatum* cyclohexadepsipeptide synthetase genes and how the structure of all these genes correlate with SEQ ID NO: 1, as well as the unpredictability of the art regarding function determination based solely on structural homology, one cannot reasonably conclude that the claimed invention is fully enabled by the teachings of the specification.

The rejections are respectfully traversed.

It is well settled that "[t]he first paragraph of section 112 requires nothing more than objective enablement. How such a teaching is set forth, either by the use of illustrative examples or by broad terminology, is of no importance." *In re Marzocchi*, 169 USPQ 367, 369 (CCPA 1971). Moreover, "a specification disclosure which contains a teaching of the manner and process of making and using the invention in terms which correspond in scope to those used in describing and defining the subject matter sought to be patented must be taken as in compliance with the enabling requirement of the first paragraph of section 112 unless there is reason to doubt the objective truth of the statements contained therein which must be relied on for enabling support." *In re Marzocchi*, 169 USPQ at 369.

"The determination of what constitutes undue experimentation in a given case requires the application of a standard of reasonableness, having due regard for the nature of the invention and the state of the art ... The test is not quantitative, since a considerable amount of experimentation is permissible, if it is merely routine, or if the specification in question provides a reasonable amount of guidance with respect to the direction in which the experimentation should proceed to enable the determination of how to practice a desired embodiment of the invention claimed ..." *Ex parte Jackson*, 217 U.S.P.Q. 804 (Bd. Pat. App. 1982).

It is also well settled that an assertion by the Patent Office that the enabling disclosure is not commensurate in scope with the protection sought must be supported by evidence or reasoning substantiating the doubts so expressed. *In re Dinh-Nguyen*, 181 U.S.P.Q. 46 (C.C.P.A. 1974). See also *U.S. v. Telectronics*, 8 U.S.P.Q.2d 1217 (Fed. Cir. 1988); *In re Bowen*, 181 U.S.P.Q. 48 (C.C.P.A. 1974); *Ex parte Hitzeman*, 9 U.S.P.Q.2d 1821 (BPAI 1988).

Moreover, in the absence of any evidence or apparent reason why compounds do not possess the disclosed utility, the allegation of utility in the specification must be accepted as

correct. *In re Kamal*, 158 U.S.P.Q. 320 (C.C.P.A. 1968). See also *In re Stark*, 172 U.S.P.Q. 402, 406 n. 4 (C.C.P.A. 1972) (the burden is upon the Patent Office to set forth reasonable grounds in support of its contention that a claim reads on inoperable subject matter).

The reasoning provided in the Office Action is that the specification does not establish the structural elements associated with cyclohexadepsipeptide synthetase function in the polynucleotide of SEQ m NO: I (or the polynucleotide used by Herrmann *et al.*) that are common in *Fusarium venenatum* cyclohexadepsipeptide synthetase genes. Applicant respectfully submits that this reasoning is not sufficient to render the claims nonenabled.

The claimed cyclohexadepsipeptide synthetase genes are structurally similar because they encode a cyclohexadepsipeptide synthetase having an amino acid sequence which has at least 70% identity with SEQ ID NO: 2; or a cyclohexadepsipeptide synthetase which is encoded by a nucleic acid which hybridizes under at least medium stringency conditions with (i) the nucleic acid of SEQ ID NO: 1, (ii) the cDNA of SEQ ID NO: 1, or (iii) a complete complementary strand of (i) or (ii), wherein medium stringency conditions are defined as prehybridization and hybridization at 45°C in 5X SSPE, 0.3% SDS, 200 μg/ml sheared and denatured salmon sperm DNA, and 35% formamide and washing three times each for 15 minutes using 2X SSC, 0.2% SDS at 55°C. One of ordinary skill in the art would, therefore, expect that the claimed nucleic acids encoding polypeptides have cyclohexadepsipeptide synthetase activity.

Furthermore, the specification contains an extensive disclosure of techniques which are well known in the art and indeed routine for persons of ordinary skill in the art for identifying other cyclohexadepsipeptide synthetase genes and disrupting or deleting such genes using Applicant's disclosure. Applicant describes methods for preparing and probing DNA libraries (Example 3); for isolating nucleic acids encoding the cyclohexadepsipeptide synthetases (Example 4); for determining cross-hybridization of the nucleic acids encoding cyclohexadepsipeptide synthetases using (i) the nucleic acid of SEQ ID NO: 1, (ii) the cDNA of SEQ ID NO: 1, or (iii) a complete complementary strand of (i) or (ii) (page 20, line 20, to page 22, line 27, and Example 2); for comparing the percent identity of the deduced amino acid sequences of the cyclohexadepsipeptide synthetases to SEQ ID NO: 2 using the Clustal method according to Higgins, 1989, *CABIOS* 5: 151-153 (page 18, line 21, to page 19, line 4); and for disrupting a gene encoding a cyclohexadepsipeptide synthetase (page 5, line 14, to page 8, line 21, and Examples 5 and 6). It is well within the skill of the art to isolate and identify the cyclohexadepsipeptide synthetase genes and disrupt or delete such genes using Applicant's disclosure.

For the foregoing reasons, Applicant submits that the new claims overcome the rejections under 35 U.S.C. § 112, first paragraph. Applicant respectfully requests reconsideration and withdrawal of the rejection.

IV. Conclusion

In view of the above, it is respectfully submitted that all claims are in condition for allowance. Early action to that end is respectfully requested. The Examiner is hereby invited to contact the undersigned by telephone if there are any questions concerning this amendment or application.

Respectfully submitted,

Date: July 7, 2004

Robert L. Starnes, Ph.D.
Reg. No. 41,324
Novozymes Biotech, Inc.
1445 Drew Avenue
Davis, CA 95616
(530) 757-8100